

SCIENCE

Some Recent Developments in the Field of Electron Microscopy

Ralph W. G. Wyckoff

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Some Recent Developments in the Field of Electron Microscopy

Ralph W. G. Wyckoff

National Institute of Health, Bethesda, Maryland

IT IS A COMMONPLACE TO SAY that experimental science is the systematic observation of nature and the attempt to set the observations thus made into an interrelated and logically consistent scheme. For this, contact with the outside world is through our senses, with vision bearing the overwhelming burden. When, in its early days, physics was concerned with the properties and behavior of matter in bulk, the unaided human eye was usually an adequate instrument. But the moment science began to occupy itself with the fine structure of matter, increase in knowledge became intimately geared to our ability to design and build instruments that would supplement, enhance, and refine our perceptions. It was thus inevitable that in penetrating more and more deeply into the structure of matter the path between what is finally perceived and the phenomenon of nature that initiated the perception should become more tortuous and the intellectual chain involved in its interpretation more complex. While it is undoubtedly a great intellectual satisfaction to devise and operate successfully such complex approaches to the secrets of nature, we sometimes lose sight of the fact that this complexity is not in itself desirable, that it inevitably limits the range and clarity of our view, and that information derived in a circuitous fashion is always come upon the hard way and is increasingly subject to error. Therefore, anything that cuts the path between what is seen and what originates the perception is bound to broaden and simplify our understanding. The electron microscope is of the greatest importance simply because it has made unexpectedly direct our perception of a world of things which we have felt certain existed but knew to be too small ever to be seen with any form of light.

It is hard to realize the great extent of the micro-world thus opened up. Several investigators have

worked for a generation in developing an ultraviolet microscopy that cannot reveal particles smaller than half those to be seen with visible light. In theory, the electron microscope can delineate particles a thousand times smaller; and, though they are scarcely 10 years old, such microscopes have already extended our vision a hundredfold. In taking this step we span that range of organized matter which extends from the animate to the lifeless and which must be understood as a basis of what we intuitively mean by "living"; at the same time we acquire the ability to "see" the larger of the molecules that are the basic units of chemistry. It rarely happens that any new experimental technique allows as direct an approach to the problems of a single science as the electron microscope thus gives to the fundamentals of both chemistry and biology.

PROBLEMS OF ELECTRON MICROSCOPY

The problems of a developing electron microscopy are threefold. The first is concerned with the design of electron lenses and their combination to produce the best attainable microscopes. The second involves a careful and thorough examination of the fine structure of biological and chemical substances with these instruments. The third set of problems deals with the application of knowledge gained in this fashion to such practical matters as the size of the submicroscopic particles in smokes, pigments, and mine dusts, or the relationships between structure and properties in materials as diverse as metallic alloys and natural and synthetic fibers. The discussion that follows is devoted mainly to the second group of problems.

Those interested in the electron microscope as an approach to the microstructure of matter need have a less intimate preoccupation with its development than was the case a few years ago. Early experimentation with a variety of designs—for emission microscopes and scattered-electron, including dark-field, microscopes and for microscopy at various volt-

Paper read at the evening meeting of the Electron Microscope Society of America, Princeton, New Jersey, 30 November 1945.

ages, to choose a few examples—has served to indicate pretty clearly the basic requirements of the kind of instrument needed to meet present problems. Also, commercial microscopes are already available which are far better than we could build if we had the facilities and inclination to try to do so. This does not mean that the existing instruments are ideal or utilize to the full the theoretical potentialities of microscopy with electrons, for it is inconceivable that an instrument so new and complicated will not undergo large-scale and perhaps revolutionary changes in design and construction as we learn better how to use it, what its rewarding uses are, and how best to make its lenses. It will indeed be a misfortune if such changes do not come about. But for those who are microscopists rather than specialists in electron optics, the important thing is that there is ready at hand an instrument whose possibilities have as yet been little explored. Too often scientific instruments become commercially available only after their most important potentialities for research have been exploited.

TYPES OF ELECTRON-MICROSCOPIC OBJECTS

The fundamental properties of electrons place certain restrictions upon what can profitably be examined in the electron microscope. Preparations must be looked at *in vacuo*, and they must be unusually thin. With this in mind, it is convenient to group objects for electron microscopy into three general classes: (1) sections cut from tissues and other materials; (2) ultrathin replicas of the surfaces of thick objects; and (3) suspensions of small cellular elements and of particles of colloidal and macromolecular dimensions. Each of these has its own problems of technique, and electron microscopy as a whole is still in that early stage of development where both the direction and the rapidity of its growth are determined largely by the evolution of these techniques.

Electron Microscopy of Tissues

Many promising biological and medical applications of the electron microscope depend on the satisfactory development of ways of cutting and handling the thin sections referred to under (1) and of differential stains for the sections thus prepared. For their micrographs to be valuable, biological sections must be less than .5 micron thick; sections from heavier materials will have to be still thinner. In the hands of very experienced operators the best available microtomes will cut down to .5 micron, but this involves pushing both instrument and skill to such a degree that other and novel procedures are obviously called for. The first to be suggested, by Von Ardenne, involved cutting wedge-shaped, instead of the usual flat,

sections. He showed that this could be done and published illustrative pictures. This method appears to have been little used in Germany, and a trial along similar lines by Richards, Anderson, and Hance has not been followed up. In this country most of the emphasis has been put on the development of a microtome to cut ultrathin sections. Such an instrument as suggested by O'Brien and McKinley and made in usable form by Ladd and Braendle and by Fullam and Gessler, is characterized by having a thin, instead of the usual massive, cutting blade put at the rapidly moving periphery of a rotor. It permits a totally new approach to many problems of cellular structure and is of the greatest importance for the future of electron microscopy. An enormous amount of careful work will be required to recognize many of the structures in sectioned tissue and to provide adequate interpretations of what is seen. Nevertheless, there are important problems that can be approached with assurance of immediately suggestive results. Thus, in spite of the discouraging early micrographs of sectioned muscle, the beautiful recent photographs of isolated tendon and muscle fibers by Schmitt, Jakus, and Hall demonstrate the extraordinary internal regularity that exists and that undoubtedly can be made apparent in satisfactorily sectioned tissues. Section through other fibrous and elastic materials, such as natural and synthetic rubbers, and plastics, will inevitably throw light on both their inner structure and what happens during their formation. It is hoped that the electron microscope will eventually make clear the kind of organization that must be present within the nuclei of cells. Attempts to observe chromosomal structure have not yet been rewarding, but progress in this direction should follow their photography in section. Another problem that can now be attacked with great profit concerns the morphology and manner of growth of viruses within the cells they infect. Most virus preparations thus far examined with the electron microscope have been purified, in the centrifuge or otherwise. A knowledge of their morphology under these circumstances is a necessary but not a sufficient step, since procedures that purify are by their nature selective and ones that separate virus from the matrix in which it is produced. The electron micrography of sections through virus-infected cells must lead to a rapid increase in our understanding of how their particles are produced.

Stereoscopic microscopy will be especially helpful in bringing out the spatial relationships between details seen in sectioned material. Since its introduction in 1940 this technique has been used with striking results upon such diverse objects as small crystal bacteria, diatoms, the wings and tracheae of insects and surface replicas of metallographic preparation

and of teeth. The ingenious reproductions of these stereoscopic micrographs, using polaroid, are especially useful for teaching and demonstration purposes.

A recent paper by Porter, Claude, and Fullam introduces another promising way of studying tissue cells. This involves their growth in culture under conditions giving single-cell spreads thin enough for direct electron photography. The pictures already published show that, where such extended sheets of cells can be grown, they yield excellent and instructive micrographs. This is another technique applicable to the problem of how viruses grow and multiply.

Electron Microscopy of Surfaces

Several ways have been devised for the electron-micrographic examination of surfaces. In early work the surface being studied was made a source of electrons which were condensed into an "image" of the surface. Depending on the material being investigated, the electrons could be either photo- or thermally emitted, or they could be electrons from a secondary source after differential scattering from the surface in question. Though these procedures have been, and will continue to be, helpful in the solution of such special problems as the electron emissivity of tungsten or oxide-coated surfaces, they have an obviously limited applicability. Closely related to the scattered-electron type of microscopy is the "dark-field" microscopy which forms images from the electrons scattered on transmission through a specimen. The German literature contains many "dark-field" pictures. Some are striking in appearance, but they do not seem to give as much information as corresponding bright-field photographs, and it is not certain how much use can be made of them.

The fine structure of the surfaces of many solids can be investigated by preparing replicas thin enough for transmission microscopy. The first replicas of this sort were described by Mahl and by Koch and Lehmann in 1940. In this country replica work was initiated by Zworykin and Ramberg. Early replicas were made of shellac, collodion, and other plastics, of silver and beryllium, and of oxide produced by anodizing an aluminum surface. The polystyrene-silica method of Heidenreich and Peck and the use of formvar by Schaefer and Harker advanced the art by providing convenient ways to study surfaces without damaging them. Formvar and other simple plastic replicas produce relatively little contrast in their micrographs and therefore reveal little of the fine detail they may contain. More contrast is obtained by the polystyrene-silica method, but these replicas too have certain limitations: (1) Since a polystyrene cast is formed at elevated temperatures and pressures, it

has a restricted application to many biological materials or to other objects whose position, form, or composition interferes with making such pressure casts; and (2), though silica gives better contrast, its apparent migration after deposition often makes equivocal the interpretation of fine detail.

Collodion replicas have not been much used until lately because little sharply contrasting detail can be seen directly on them. It has generally been assumed that this is due to the fact that collodion does not give a faithful small-scale reproduction of surface detail. Metal shadowing, as developed over the last two years by Williams and Wyckoff, has indicated that this is not the case but that, on the contrary, properly made collodion or formvar replicas reproduce fine structure all the way down to molecular dimensions. Since plastic replicas formed from solution are especially easy to prepare and can be taken from fragile and relatively inaccessible surfaces, and since metal shadowing brings out the detail on them, such shadowed collodion or formvar replicas are applicable to many problems. Evidently techniques are now at hand for teaching much that is new about the surfaces of metals, etched for metallographic analysis, machined in various ways, corroded, or worn by use; of glass and ceramics during and after a variety of chemical and physical treatments; of crystals found in nature or prepared in the laboratory; of many biological structures, such as large cells, microorganisms, and teeth; as well as of macromolecules distributed over a relatively smooth surface.

Electron Microscopy of Particulate Suspensions

To many, the overwhelming attraction of the electron microscope lies in the unique opportunity it offers for actually seeing big molecules. Partly for this reason and partly because relatively simple techniques of observation are required, much effort has already gone into the study of suspensions of small and submicroscopic objects. Information has been sought about the size distribution in many inorganic colloidal materials, such as gold and other metallic sols, clays, carbons, pigments, dusts, and smokes, and many practically helpful results have come out of this work.

Suspensions of microorganisms, like colloids, have been the subjects for numerous electron micrographs. Bacteria were among the first objects to be examined, a first photograph having been published by Marton as early as 1937. The morphological survey of various bacterial species which has followed, and which is as yet far from complete, is a necessary preliminary to other uses of the electron microscope by bacteriologists. Some have expressed disappointment with

what the microscope has shown about bacteria, and there have indeed been no exciting discoveries about the fundamental properties of their nuclear and protoplasmic structures. Perhaps the inevitable improvements that will come about in ways of handling and preparing these relatively large objects will sometime give sensationally interesting results, but whether this happens or not, sound morphological studies, repeated if necessary with each advance in technique, are necessary. This is equally true for other microorganisms, both larger and smaller than bacteria. There have been investigations of spirochetes, such as those of syphilis and infectious jaundice; but serious studies of many other interesting large microorganisms, such as molds and the malarial parasite, remain to be made. Nor has much yet been published about the especially small organisms, like the highly pleomorphic causative agent of pleuropneumonia or the infectious units of trachoma, psittacosis, and related diseases, that lie between bacteria and viruses. Their very minuteness, which renders observation by classical methods so difficult, makes them particularly desirable objects for electron microscopy.

The electron microscope offers a new way to investigate the chemical and immunological reactions of bacteria and bacterial products. Except for photographs of the capsules of pneumococci and of bacterial cellulose, the products of bacterial metabolism remain to be investigated; and only preliminary papers have appeared dealing with the potentially fruitful field of the mechanism of the action of antibiotics and germicides on bacteria. Electron microscopy is a direct approach to the mechanism of the reaction that occurs between an antigen and its antibody. Here again, existing work is of a preliminary character, being restricted mainly to a demonstration that the reaction is discernible in electron-microscopic preparations. Thus, Mudd and Anderson have shown the change in appearance of typhoid and other bacteria in the presence of their antisubstances, the apparent thickening of flagella by flagellar antibodies, and the capsular swelling that takes place when a pneumococcus is mixed with its antibody. For the improvement of vaccine and serum production, as well as for more fundamental reasons, it is important to separate and purify from one another the various antigens associated with a bacterium. The ability of the electron microscope to make evident macromolecules means that the larger of these antigenic components can be photographed; Shepard and Wyckoff have in this way "seen" what is presumably the soluble antigen of typhus and its reaction with antirickettsial serum. This method of study can be applied to many problems in bacteriology.

A most interesting group of isolated objects larger

than bacteria are not microorganisms but suspensions of cells and tissue elements, of both plants and animals. In connection with a study of the clotting of blood, early photographs of platelets and of the walls of erythrocytes were published. Claude and Fullam have photographed spherical elements from the cytoplasm of leukemic cells from the rat which they considered to be mitochondria. Preliminary pictures have also been taken, both in this country and abroad, of the analogous chloroplasts from plant leaf cells. Spermatozoa have been photographed and their tails found to have a fibrillar structure. An early examination was made of the fine details of bird feathers and fragments of the chitinous shells of insects. Especially suggestive for further work is the observation of a fine structure, resembling that of collagen, in photographs of iridescent insect scales. But this was not found in nerve axoplasm of the squid, nor has it been seen yet in isolated chromosomes and other nuclear material from cells.

The most impressive demonstration thus far of structure within tissue elements results from the study by Schmitt and his co-workers of separated phosphotungstic acid-stained fibers of tendon and the correlation of this electron-microscopic structure with the X-ray diffraction effects it produces. This regularity independent of width, is seen in fibers whose cross-section grades downwards to the limit of visibility with present-day electron microscopes. Evidently we are dealing here with elements that approach the fibrous protein units. Electron-microscopically visible structure will doubtless be found in other fibrous proteins. There is evidence for structure in fibrin, and it is clear in the micrographs of separated muscle fibers and trichocysts of paramecia. Only a start has been made in the electron-microscopic analysis of wood and cellulose.

LIMITS OF ELECTRON-MICROSCOPIC VISION

Two factors dominate the photography of especially small particles and minute details of structure: contrast and resolving power. As already stated, the limit of resolution of existing microscopes is approached only when dealing with particles of molecular dimensions. Contrast is rarely a problem with photographing smokes, dusts, and elastics like rubber that do not have to be supported on membranes, but it is often a limiting factor in the photography of inorganic colloids. But macromolecules are organic and hence are composed of light atoms whose scattering power for electrons is low. There is so little matter in these molecules that, especially after being mounted on supporting membranes, they offer very slight contrast; in practice the lower limit of what can be seen clearly is set by such lack of contrast

rather than by insufficient resolving power in the microscope. For the satisfactory observation of macromolecules and of molecular detail in organic material it is therefore necessary to have recourse to a staining or impregnation that will enhance contrast. Two ways of doing this suggest themselves. One, to bring out internal structure, is analogous to the customary staining of biological material except that a "stain" for electrons is a substance of great scattering power. Such chemical staining was involved in the treatment of collagen with phosphotungstic acid; it occurs when this or some other protein coagulant—or a relatively heavy ion, for instance—is added to a virus preparation. Williams and Wyckoff have found that metal shadowing, involving the oblique deposition of heavy atoms that do not subsequently migrate over the preparation, also acts as a surface stain to increase the visibility of macromolecules and bring out their shapes. Müller, in Germany, like ourselves, tried the oblique evaporation of metal as a device for measuring the heights of electron-microscopic objects; but he worked with silver which does not give the continuous, essentially grainless, film needed to show fine detail and to cause the three-dimensional impression we have found useful in outlining particle size and shape. Mahl also tried evaporating chromium onto replicas, but there does not appear to have been any follow-up of this type of work in Germany.

At present it is impossible to say exactly what is the smallest particle that can be recorded clearly in electron micrographs. This question is obviously of vital concern to all who are occupied with the photography of molecules. Resolving power of an optical system is commonly defined in terms of its ability to show as separate two objects exceedingly close to one another. A picture published by Von Ardenne contains two particles of colloidal gold separated by not more than 40 Å.; in another photograph, by Von Borries and Ruska, inorganic particles appear separate, though the distance between their centers cannot exceed 25 Å. Von Ardenne has called attention to an image which he ascribes to a particle no more than 10 Å. across—apparently the smallest anyone has yet claimed to have seen with the electron microscope. In studying molecules one is more interested in the ability of the microscope to provide a faithful image of a molecule than in mere indications of its presence. It is not simple to tie this up with such a definition of resolution as the foregoing, but more will be known about this after photographs have been made of several molecules of established shape. Even if the foregoing represents a somewhat optimistic estimate of the powers of the average microscope, we are now in a position to investigate most proteins, both globular and fibrous; many synthetic long-chain polymers;

polysaccharides including dextrans, starches, and cellulose; and the world of viruses that reaches upward to the lower limit of visibility of the optical microscope. With the means now at our disposal it is altogether probable that many new molecular entities will be discovered within this region.

PHOTOGRAPHY OF MACROMOLECULES AND OF VIRUSES

Attempts to photograph macromolecules began early in the history of electron microscopy. The first molecule-like particles to be recorded were tobacco mosaic fibrils, which Kausche and Ruska showed to be the elongated fibers now familiar to all. This and other early work on plant viruses by the German school was followed by photographs of molecular suspensions of hemocyanin, edestin, glycogen, and an iodine-containing reaction product, *p*-iodobenzoyl glycogen. Pictures of hemocyanin and edestin molecules, by Stanley and Anderson, were the first to appear in this country. Since then, excluding studies of rubber, collagen, and polymers, whose fibers are not molecules in the sense used here, there has been only a limited extension of molecular photography.

Williams and Wyckoff have recently shown that metal shadowing permits considerably improved molecular photography; and the practical lower limit of size of molecules that can be distinguished is now set by the particulate structure of the collodion or other substrate. To work with the smaller molecules, then, this collodion structure must be minimized or a smoother substrate found. Formvar is not much smoother, but we have found that its structure or that of collodion can be largely suppressed through recourse to a replica technique wherein molecules on a smooth glass surface are shadowed and the resulting film backed up with unshadowed plastic. Such replicas have already delineated molecular particles at least as small as 100 Å.

These experiments should be extended in two obvious directions. One is concerned with photography of smaller and smaller molecules; the other involves photography of the products of reactions between macromolecules or of the substitution of heavy atoms into such molecules. The reaction between an antigen and its antibody is the best known, and certainly one of the most important, intermacromolecular reactions; others deal with the adsorption of colloidal particles onto macromolecules, and the polysaccharide-protein, lipid-protein, and nucleic acid-protein combinations that are so common in nature. It would be rash to try to foretell what the photography of macromolecular reaction products will show, but obviously we have a new way of examining the denaturation of proteins and their change in molecular shape and size with changes in pH and under the action of such

splitting agents as urea; and it is not unreasonable to look forward to the time when we will be able to see directly the action of enzymes.

Shadowed molecular photography permits study of the structure of collodion and other reaction and split products of cellulose. In a similar way information about muscle will come from the photography of its extracted protein myosin. Such investigations of chemical derivatives will undoubtedly take their place alongside studies of mechanically disintegrated and sectioned material as approaches to the structure of fibrous substances.

Just as viruses are intermediate in size between molecules and microorganisms, so the problems they present are partly molecular and partly biological. Not only can the electron microscope define the sizes and shapes of elementary virus particles, but it also offers a way of examining details of the chemical reactions into which they enter and of seeking how they originate within their host cells.

Electron micrographs made of purified viruses show particles whose dimensions are in general agreement with the results of indirect physicochemical measurements. Through the work of Krause, Stanley and Anderson, Sharp, and others we now have micrographs of several plant viruses, the eastern and western forms of encephalomyelitis, the rabbit papilloma, several strains of influenza, vaccinia, and ectromelia. Photographs have also been made of infectious material from hoof-and-mouth disease, silkworm jaundice, human and murine poliomyelitis, chicken pox, and herpes. Partly because of the unusual sperm-like shape of the particles of some strains, bacteriophages have proved excellent material for electron microscopy.

The particles in some of the purified virus preparations are uniform in size; with others, size varies about a mean. Thus, the spherical particles of the bushy stunt plant and the papilloma animal viruses appear alike, their diameters being those already determined through ultracentrifugation. The spherical particles of such a virus as influenza, on the other hand, are far from uniform as seen under the microscope. It is sometimes suggested that such variation is evidence for a microorganismal, as opposed to an essentially molecular, character, but this is not a valid argument since similar variations in measured size occur among the molecules of a protein like hemocyanin. This nonuniformity in diameters presents an important problem. It may indicate that the molecules of a protein need not be all alike, or it may be a consequence of an irregular shrinkage or desiccation of particles strongly hydrated in solution. This will be answered when we have enough accurately measured micrographs of several kinds of molecules and

a correlation of these measurements with particle size and degrees of hydration as determined by physicochemical analysis.

Important chemical reactions of viruses are those in which they are inactivated, as for vaccine production or are neutralized by specific antiserum. There has been no thorough microscopic study of inactivation, though treatment with formaldehyde is known to have little, if any, effect on the appearance of the elementary particles of some viruses. A splitting of the tobacco mosaic fibers by physical means has been photographed. Micrographs already made show that the reaction between virus and antibody is readily recognized: the virus particles appear bigger and more diffuse in outline, presumably due to attached antibody, and agglutination on a microscale is common. If certain current hypotheses are correct in suggesting that this combination involves the formation of antigen-antibody lattices, such regularity should be clearly evident. On the practical side the minute amounts of virus, or antibody, that go to make up a microfloc indicate that highly sensitive diagnostic tests can be built around the electron-microscopic observation of this immune reaction. Another reaction, chemical or biological, upon which microscopic studies have already been begun, is the lysis of bacteria by the bacteriophages.

The electron microscopy of crystallizable viruses and proteins offers a direct approach to questions of how crystals are built up from their molecular units. Crystal formation can be photographed, and this will provide very precise knowledge of many of the factors that determine crystallization, of how microcrystals form and grow, and of how true crystallinity is related to the paracrystallinity observed with fibrous materials like the tobacco mosaic virus.

The problem of how the elementary particles of a virus come into being can now be investigated. Once these particles have been identified through work with purified suspensions, the road is open for dealing with less pure preparations, with the ultimate object of seeing viruses in the cellular environment in which they develop. Only a first step has been taken in this direction, but it has been recognized that filaments as well as spheres are present in purified influenza virus suspensions and that an intimate relation seems to exist between the two forms. Thus, we now have a way to see how viruses grow; and it is possible that these experimental procedures may ultimately tell us how large "inanimate" molecules, like those of hemocyanin, originate. It is worth reiterating that with the electron microscope as guide and tool we are entering a world where, very literally, life begins. What will be found there, beyond useful information about viruses and the diseases they cause, can scarcely be foretold.

Technical Papers

Radiochemical Changes in Some Fatty Acids¹

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Since recent marine sediments are believed to be a possible—even likely—starting point in the formation of petroleum, some of the solid and semisolid organic compounds contained in such sediments have been investigated. Among these compounds are fatty acids, whose presence in recent marine sediments is well established, even though it is not known in all cases whether they occur in the form of fats, metallic salts, or free acids. For this reason the radiochemical decomposition and conversion of a number of fatty acids and their salts have been studied.

The data are most complete for the following three long-chain fatty acids: caprylic ($C_7H_{15}COOH$), lauric ($C_{11}H_{23}COOH$), and palmitic ($C_{15}H_{31}COOH$). They were bombarded with alpha particles from radon and with deuterons from the M.I.T. cyclotron. All three of these acids are members of the same family, and are made up of a straight paraffinic chain or aliphatic radical, R, of varying length, joined by a C-C bond to the carboxyl group, $COOH$, characteristic of the organic acids. The alpha-particle bombardments were carried out by coating a 4-gram sample of acid, which is solid at room temperature, on the inside of a glass bulb, which was then evacuated. Next, about 100 millicuries of radon were admitted and allowed to form an active deposit on the acid coating. The course of the reaction was followed by reading on a manometer the pressure of gases produced. Deuteron bombardments were made in an evacuated, water-cooled, metal chamber, which admitted the cyclotron beam through a copper foil and allowed the gases produced to be collected. After a period of time sufficient to allow the radon activity (for alpha runs) or the various induced copper activities (for deuteron runs) to drop to a safe level, the gaseous products were analyzed and an attempt made to identify some components of the complex liquid-solid mixture.

¹Talk presented at the Springfield meeting of the New England Section of the American Physical Society, 13 October 1945.

During the past three years, the effects of terrestrial radioactivity on the genesis of petroleum have been studied under American Petroleum Institute Project 43c. This project, entitled "Transformation of Organic Material into Petroleum—Physical and Physicochemical Phases," is under the direction of Profs. W. J. Mead, Clark Goodman, and W. L. Whitehead at M.I.T. Most of the alpha-particle bombardments and chemical analyses were done by Dr. C. W. Sheppard, who was a member of the group until the Summer of 1945. He was assisted by Miss Virginia Burton. Some of the mass spectrometric analyses were performed by Mr. E. C. Farmer.

Small amounts of lower fatty acids, which are water soluble, were isolated by water extraction. The most important liquid product, isolated by microdistillation under high vacuum, could be identified by employing every method of physical and chemical analysis available on a microscale. This was pentadecane, $C_{15}H_{32}$, from palmitic acid and undecane, $C_{11}H_{24}$, from lauric acid (1). Thus, the straight-chain hydrocarbons just mentioned must have been produced by the rupture of the R-COOH bond and the addition of an H atom to the long-chain radical. The yield of these liquid products was approximately 80 mg./100 millicuries in both cases. Finally, small amounts of an amber, vaseline-like residue were left over after the unconverted original acid was changed to its sodium salt and removed. The complex liquid-solid mixture from the caprylic acid bombardment was not analyzed.

The radiochemical decomposition and conversion of the three fatty acids investigated was further studied

TABLE 1
ANALYSES OF GASEOUS PRODUCTS FROM ALPHA-PARTICLE BOMBARDMENTS OF FATTY ACIDS

Acid	Mole per cent of constituents									
	CH_4	C_2H_6	C_3H_8	C_4H_{10}	H_2O	H_2	CO	CO_2	$CO + CO_2$	$\frac{H_2}{CO + CO_2}$
Caprylic $C_7H_{15}COOH$	0.7	1.0	0.4	0.7	3	33	10	51	61	0.54
Lauric $C_{11}H_{23}COOH$	0.6	0.5	0.1	0.2	4	42	11	41	52	0.81
Palmitic $C_{15}H_{31}COOH$	0.4	0.6	0.1	0.8	10	48	6	34	40	1.20

with the help of the gases produced. The gas mixtures were analyzed on a small Nier-type 60° mass spectrometer which had been developed for this particular purpose. The results obtained were checked by analyses performed by a low-temperature condensation and absorption method. While this latter method seems to be most reliable for the determination of CO_2 and H_2 , which were always present in large quantities, the mass spectrometer is far superior for the positive identification of small amounts of hydrocarbons. Table 1 gives the mole per cent of constituents in the gas phase and represents the weighted mean of mass spectrometer and low-temperature values. It is seen that in every case the main constituents are CO_2 and H_2 . CO and H_2O are found in moderate amounts, while the concentration of the paraffins present is low.

CO_2 and CO can be formed directly from the

carboxyl group, COOH , when this group breaks up into fragments after having been severed from the original acid molecule by R-COOH bond rupture. The balance of these fragments, *i.e.* the OH groups and H atoms, may contribute to the formation of H_2O and H_2 . Simple computations show, however, that this takes care only of part of the large amounts of H_2 formed. Thus, most of the H_2 must derive from H atoms produced by C-H bond rupture. The formation of the light paraffins, CH_4 to C_4H_{10} , is most readily explained by C-C bond ruptures near the

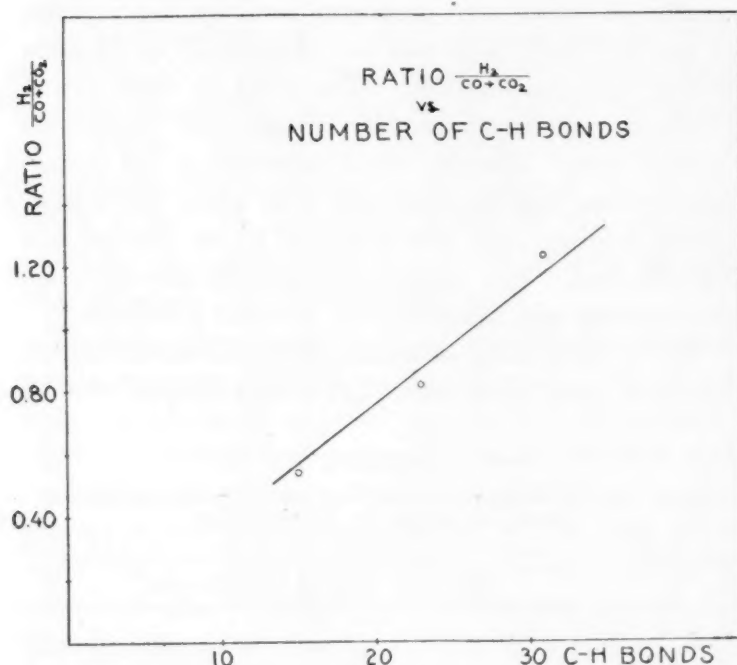


FIG. 1. Abundance of hydrogen relative to carbon monoxide plus carbon dioxide as a function of the number of C-H bonds in the molecule.

"left" end of the molecules, yielding methyl to butyl radicals. Free H atoms can combine with these radicals and produce the corresponding paraffins. These simplified considerations do not include the possibility that part of the gases found in the analysis may derive from reconverted primary gases.

In the case of palmitic acid, for which the available data are most complete, it is possible to perform computations which give a rough idea of the statistical probability of the rupture of different bonds in this molecule under alpha-particle or deuteron bombardment. Thus, it is found that, for every 100 C-H bonds broken, about 45 C-C bond are ruptured at the junction of the long chain and the COOH group, while probably not more than 5 C-C bonds are broken anywhere else along the chain. These ratios are of interest because they give an indication of bond strengths. Since at least 90 per cent of all the C-C bond ruptures seem to occur at the terminal COOH group, the strength of this particular bond must be considerably lower than that of the paraffinic C-C bonds (about 80 kcal./mole). This conclusion is con-

firmed by considering the ratio of R-COOH and C-H bond ruptures, which is far in excess of estimates just based on the relative number of these bonds available.

A comparison of the actual gas yields from the three acids cannot be carried out, since some of the data necessary are not sufficiently accurate. However, if for the three acids investigated the abundance of H_2 relative to CO plus CO_2 is plotted *vs.* the number of C-H bonds in the molecule, a straight line is obtained (Fig. 1). Thus, the assumption seems justified that in long-chain molecules the probability of breaking C-H bonds is directly proportional to their number.

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Surface Phagocytosis—Its Relation to the Mechanism of Recovery in Pneumococcal Pneumonia¹

W. BARRY WOOD, JR., MARY RUTH SMITH,²
and BARBARA WATSON²

During the course of a systematic study of the effect of chemotherapy upon the pulmonary lesion of pneumococcal pneumonia it has been shown that pneumococci are destroyed in the lung by phagocytosis (8, 9) and that the phagocytic process takes place in the absence of demonstrable circulating antibody (11). All existing experimental evidence indicates that virulent pneumococci are protected, by virtue of their capsules, against the attacks of phagocytic cells (2). Both polymorphonuclear leucocytes and macrophages have been found to ingest fully encapsulated pneumococci only in the presence of type-specific opsonin (6). Thus, the phagocytic reaction which takes place in the lung during chemotherapy remains unexplained.

Three possible explanations may be offered for the occurrence of phagocytosis in the absence of demonstrable circulating antibody: (1) The reaction may be brought about by the local accumulation of specific antibody in the pneumonic lesion; (2) it may result from injury to the pneumococcus capsule; or (3) it may be due to a hitherto undescribed mechanism which is related neither to opsonins nor to capsular injury. Although all attempts to substantiate the first and second of these hypotheses were uniformly unsuccessful (11), the third explanation was conclusively confirmed by direct observation (12, 13).

¹ From the Department of Medicine, Washington University School of Medicine and the Oscar Johnson Institute for Medical Research, St. Louis, Missouri. These studies were supported by a grant from the Commonwealth Fund.

² Recipients of Research Fellowships awarded by the Lederle Laboratories, Inc.

Suspensions of thrice-washed phagocytic cells³ in gelatin-Locke's solution were mixed with washed pneumococci (Type I, Strain A5), and the mixtures were kept on ice until used for phagocytic tests. In successive experiments the phagocyte-pneumococcus mixtures were injected intrabronchially into (a) the lungs of normal rats, (b) lungs removed from rats and perfused with gelatin-Locke's solution, and (c) rat lungs fixed for 24 hours in 10 per cent formalin and washed for several days to remove the fixative. Each experiment was carried out at body temperature. Sections cut from all three types of preparations and stained by the Gram-Wiegert technique showed clearly that both polymorphonuclear leucocytes and macrophages phagocytized pneumococci in the alveoli within less than an hour. In the experiments with the formalin-fixed lungs, there was no possible source of intermediary opsonin.

Further examination of the formalin-fixed lungs revealed that pneumococci were engulfed by the phagocytic cells in the large bronchi as well as by those in the alveoli. Phagocytosis failed to occur, however, when the same leucocyte-pneumococcus mixtures were tested in rotating glass tubes, hanging-drop preparations, and even in capillary tubes of the same diameter as the bronchi. This last finding suggested that the crucial factor in the phagocytic process was related in some way to the character of the bronchial surface.

Other tissue surfaces were therefore tested. Small pieces of selected tissues taken from freshly killed rats were placed in the bottom of Petri dishes lined with moistened filter paper to prevent drying during incubation. A small drop of the leucocyte-pneumococcus mixture was spread over each tissue surface, and the Petri dishes were then closed, sealed with Scotch tape, and incubated for one hour. At the end of incubation, impression smears were made of the tissue surfaces and stained with methylene blue. Bronchial and tracheal epithelium, esophageal epithelium, the intima of both aorta and vena cava, lung, pleura, pericardium, endocardium, peritoneum, liver, spleen, kidney, mesentery, retina, muscle, and clotted plasma all were found to support phagocytosis. When the tissues had been boiled previously for 30 minutes the phagocytic reaction still took place. Finally, a variety of inert surfaces were tested, and although phagocytosis failed to occur on glass, ground glass, paraffin, albumen, and cellophane, phagocytes brought into contact with filter paper, blotting paper, lens paper, cloth, and fiber glass were found to be highly active.

From these observations it may be concluded (1)

³The phagocytes were obtained from peritoneal exudate of rats inoculated 24 hours previously with an aleuronat-broth mixture. Approximately 90 per cent of the cells were polymorphonuclear leucocytes, and 10 per cent, macrophages.

that both polymorphonuclear leucocytes and macrophages, when given access to a suitable surface, will phagocytize virulent pneumococci without the aid of an intermediary antibody or any other tissue factor, and (2) that most body tissues afford surfaces suitable for the efficient operation of phagocytic cells in this nonantibody reaction.

Due largely to the profound influence upon immunological thought of the now-classic investigations of Avery and his collaborators (7), most of the previous studies of the mechanism of recovery in pneumococcal pneumonia have centered about the role of specific antibodies. The methods of classical immunology, however, have failed to reveal why untreated patients sometimes recover from pneumococcal pneumonia before specific antibody is demonstrable in their blood sera (3, 5), why sulfonamide chemotherapy usually causes a crisis several days before immune bodies appear in the blood (1, 10), and why phagocytes destroy pneumococci in the lungs of patients dying of pneumonia even when the pneumonic lesion contains large quantities of unbound antiphagocytic polysaccharide (4). The phenomenon of surface phagocytosis described in this report offers an adequate answer to each of these previously unsolved questions. It also explains the phagocytosis of fully encapsulated pneumococci in experimental pneumonic lesions in the absence of specific opsonins and in the presence of excessive amounts of polysaccharide. In view of the tremendous surface area afforded by the alveolar architecture of pulmonary tissue it seems logical that surface phagocytosis should constitute an important defense against bacterial invasion of the lungs. Preliminary experiments already indicate that this nonantibody phagocytic mechanism operates in other body tissues and is responsible for the destruction of other species of encapsulated microorganisms.

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Transplantation of Adult Filarial Worms, *Litomosoides carinii*, in Cotton Rats¹

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The experimental study of filariasis is handicapped by the prolonged period of development of the filarial worm in man and other animals. Among laboratory animals the dog may be considered a suitable subject for experimental purposes, but the location of the parasite in the pulmonary artery limits observation, is too perilous for manipulation, and does not resemble that of the filarial worms inhabiting man. In recent years the finding of a filarial worm commonly occurring in the pleural and pericardial cavities of the cotton rat, *Sigmodon hispidus*, puts at our disposal an infection which resembles in many ways certain forms of human filariasis. With the view of studying some aspects of this infection, we have resorted to an unorthodox approach which consists of the transplanting of the adult worms from one definitive (*i.e.* final) host to another of the homologous species, which, with a metazoan parasite, is a departure beset with implications.

Cotton rats infected with *Litomosoides carinii* were etherized, tied to a board, and shaved over the thorax. The body was swabbed with a solution of phenol and the thoracic area washed over with alcohol. A slit was made along the midanterior line with scissors and continued along the sternum up to the clavicle. The sides were pinned down, exposing the pleural cavities, which, after this procedure, should be free of blood. The worms were withdrawn aseptically with light forceps or, better still, with a hooked rod and placed in sterile saline in several Petri dishes.

The rat into which the worms were to be transplanted was etherized, tied to the board, shaved and swabbed, and punctured with a thin blade in the central area of the right anterior thorax. By means of blunt hooks the opening was kept sufficiently large to insert 5 to 25 worms with the aid of a blunt probe and with a minimum amount of injury to the worms. The wound closed and healed without aid.

The attempt to transplant live adult *Litomosoides carinii* from the pleural cavity of an infected cotton rat into that of a normal cotton rat has so far failed in 10 animals. As early as three days following transplantation, the worms are dead and are found to be accumulated in a single mass which progressively becomes a syncytium of degenerating worms, as de-

termined from later observations with other treated animals. Live microfilariae, however, may be found for a week or longer in the pleural cavity and in the blood. There is no gross exudation of fluid into the cavity, which appears perfectly healthy, with the syncytium of worms cleanly isolated in a serous covering.

In contrast to the negative results in normal rats successful transplantation of the adult worm was accomplished in splenectomized cotton rats, the reticuloendothelial system of which had subsequently been blockaded with India ink. Live worms in variable numbers were recovered from the pleural cavity in four out of nine treated rats, with indications that in one other case the worms survived sufficiently long to migrate into the left cavity. In the course of these observations it was noted that the male worm often survived assaults of the defense mechanism of the host which destroyed the females, whose exudations appeared to be much more irritating than those of the males. Worms transplanted into one side of the thorax are later found within both cavities in approximately equal numbers, and may be seen to migrate across the mediastinum, through the fatty tissues, even between the parietal pleura and the diaphragm, emergent worms having been seen partly through these structures.

Successful transplantation has also been achieved in two out of three rats as the result of X-irradiation with 140 kv., 5 M.A. (without filters) at 25 cm., using a large cone (over 7 cm.) and delivering 120 r per minute. The type of cell and tissue most affected by this treatment, as contrasted with that of blockade, offers significant data in considering the resistance manifested by the host under normal and experimental conditions.

Success has also attended transplantation from one to another naturally infected rat. Also, rats injected four to five times in the course of as many weeks with a suspension of *Dirofilaria immitis* contained a mixture of Falba, mineral oil, and killed tubercle bacilli (1) have accommodated transplants of live adult *Litomosoides carinii*, and in one case lymph node invasion has been observed.

Pathological findings will accompany more detailed publication.

Addendum: Since this article was accepted for publication, Scott and Cross (2) have described a tumor in the superior mediastinum of old cotton rats, caused by dead adult filarial worms, and have assumed the neutrophil-infiltrated tumor to have been caused by the dead worms. In view of this observation, it has been thought advisable to note here certain general findings on the pathology of filariasis in the cotton rat which have been made in a number of animals nat-

¹This work was done with the aid of grants from the Marcelle Fleischmann Fund for the Study of Immunologic and Allergic Phenomena in Tropical and Parasitic Diseases and the American Foundation for Tropical Medicine.

rally and experimentally infected with *Litomosoides carinii*.

Scott and Cross's observation occurs not infrequently in our experience as a result of worms dying in transit through the fatty lymphoid tissue in the region of the superior mediastinum near the hilus of the lung. This is accidental. But a characteristic feature of the infection is a generalized or spotty proliferative reaction of the visceral and parietal pleura which, in its final phase, results in papillary nodulations of the superficies. Neither the worms nor the nodules invade the lung proper, though it shows reaction to the infection. The spleen is hypertrophied.

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The Effect of Thiouracil Upon Pigmentation in the Tadpole

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In the course of experimental work concerning the effects of thiourea and thiouracil¹ upon the thyroid glands of tadpoles (*Rana sylvatica*), the latter drug was found to cause pigmentary changes which seem worthy of record.

The tadpoles used in this work were obtained from eggs collected in near-by Maryland. The animals were raised in large finger bowls with 20 tadpoles in each bowl and were fed a diet of boiled spinach, Pablum, and boiled egg. Ten of the cultures served as controls, being kept in tap water; in the case of 10 others the culture fluid was a .05-per cent solution of thiourea, and that of another 10 was a .05-per cent solution of thiouracil. These treatments were started on 19 March 1945, when the tadpoles were in the tail-bud stage. The culture fluids were changed every other day. On 4 April, 16 days after the beginning of the experiment, it was noted that the animals in the thiouracil series were considerably lighter in color than were those in the other two groups. This difference became more striking, so that by 14 April (26 days) all tadpoles of the thiouracil series showed very marked blanching with the melanophores contracted in a manner similar to that seen in hypophysecto-

mized specimens. By this time the inhibiting effect of the drugs upon metamorphosis, reported by other authors (1, 2), was clearly evidenced, the controls showing rapid growth of the hind limbs, while the experimental animals of both sets had only rudimentary limb buds. Detailed results of the experiment as they relate to the thyroid and metamorphosis will be reported elsewhere. By 4 May (56 days) 80 per cent of the controls had metamorphosed while none of the experimental animals had shown any signs of metamorphosis. At this time, therefore, the treatment was discontinued and the experimental animals transferred to tap water. The thiourea-treated tadpoles did not begin to metamorphose despite discontinuance of the treatment, and on 27 July (129 days) no increase in hind-limb length had occurred in this group. This does not accord with previously reported results (1), but the difference may be related to the slightly higher concentration used in the present experiments. The animals of the thiouracil series, on the other hand, showed signs of the initiation of metamorphic changes very quickly after their removal to tap water. By 26 May (67 days) 50 per cent of these specimens exhibited definite elongation of the hind limbs, and by 11 June (83 days) 80 per cent had metamorphosed. Moreover, these specimens resumed the normal dark color quite rapidly, so that within a week after the thiouracil administration ceased, all of the tadpoles showed the same degree of expansion of the melanophores as had been seen in the controls.

It appears that thiouracil has some specific effect upon the melanophores of the tadpole and that such an effect is not produced by a similar concentration of thiourea. Work is now in progress to ascertain whether this action is exerted directly upon the melanophores or indirectly through the intermediation of the pituitary gland.

Juhn (3) has reported an effect of thiouracil upon the pigmentation of the feathers of Brown Leghorn capons, but this is apparently to be attributed to the inhibition of thyroid function by the drug, since it is similar to the pigmentary change which follows thyroidectomy in this animal.

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¹The drugs used were supplied through the courtesy of the Lederle Laboratories, Inc.

News and Notes

Carl E. Swartz, formerly chief metallurgist of the Cleveland Graphite Bronze Company, has been appointed division engineer in charge of materials research, Kellex Corporation. Dr. Swartz will be assigned to the research unit of The Johns Hopkins University Applied Physics Laboratory, 8621 Georgia Avenue, Silver Spring, Maryland, with which the Corporation has recently become associated in a special project for the U. S. Navy.

Robert L. Pendleton, soil scientist of the Office of Foreign Agricultural Relations, U. S. Department of Agriculture, has been loaned for four months to the Ministry of Agriculture and Lands, Kingdom of Siam. He is assisting his former associates in their plans for returning to a peacetime basis. The seven years he spent in Siamese Government service as soil scientist and agriculturist peculiarly qualify him for the present assignment.

George T. Scott has been made assistant professor of zoology at Oberlin College.

Benjamin McKeever has been appointed assistant professor of psychology at the University of Pittsburgh, effective in June.

Announcements

On 3 July the Senate passed S. 1850 providing for a National Science Foundation. The action followed two days of debate during which Senator H. Alexander Smith lead an attempt to replace the Kilgore-Magnuson Bill. The Social Science provisions were not included.

The American Council on Education is undertaking a country-wide survey of pharmaceutical education. The American Association of Colleges of Pharmacy initiated the proposal for an over-all study of pharmaceutical education with special reference to the modern practices and services required of pharmaceutical graduates, and funds for the survey—approximately \$100,000—will be provided by the American Foundation for Pharmaceutical Education.

The plans for the survey include a careful study of the supply of, and future demand for, trained pharmacists; the practices of the colleges of pharmacy as to the admission, guidance, selection, and training of students; analysis of present-day prescriptions to determine the knowledge required of the professionally trained pharmacist; the relation of pharmaceutical education to business and industry; the role of pharmacy in medical care; the qualifications of

faculty members and the conditions of faculty service in the colleges of pharmacy; and the relation of the requirements for licenses to the program of training and practical needs. Special attention will be given to methods and means for the training of the scientific specialist now required by the rapidly growing pharmaceutical industries and for pharmaceutical research.

The projected pharmaceutical survey will be under the direction of Edward C. Elliott, formerly president of Purdue University, who will be assisted by a technical staff. There will also be appointed an advisory committee representative of educational interests, state licensing boards, and the pharmaceutical industries. It is expected that the survey will require from two to three years.

Recent research on tryptophane (3rd ed.) has been prepared by the Special Chemicals Division of Winthrop Chemical Company, Inc., 170 Varick Street, New York City, and is now being distributed on request. The current edition brings this review of the literature up to February 1946 and includes 159 references.

Among the fields in which research on tryptophane has been most active during the past year are its interrelations with nicotinic acid and pellagra, with pyridoxine and anemia, and with bacterial and viral nutritional requirements. A striking development was the discovery that sodium acetyl-dl-tryptophane can be used effectively for fortification and stabilization of serum albumin. Chief commercial use of tryptophane remains the fortification of various protein hydrolysates.

A new opportunity for limited employment of qualified graduate students in the field of map research has been announced by the Army Map Service Corps of Engineers. The Association of American Geographers is supporting the project through a committee which will attempt to recruit and screen candidates.

This is an unusual opportunity for students in geography, enabling them to complete necessary research for a master's thesis or a doctor's dissertation and acquire practical experience, in addition to being paid for their work. It is intended that each student complete his work in a form suitable for publication and the Army Map Service will supervise his work toward that end and be responsible for the quality of the project. The approval or disapproval of the work as partial fulfillment toward a degree, however, will remain with the institution granting the degree.

The Association's committee hopes to be able to provide the Army Map Service with a list of potential applicants from which it can make selections as soon as possible after 1 June. All applications should include a brief but comprehensive résumé of the applicant's qualifications and experience, with a recommendation and appraisal of the student from some member of the faculty. This information is to be used not only by the committee in compiling its list of potential applicants, but by the Army Map Service in the final selection and orientation of the students.

The selected candidates actually become civil service employees of the Federal Government, assigned to the Army Map Service, Corps of Engineers, U. S. Army, in Washington, D. C. As such, they are subject to the hours, regulations, and benefits of Civil Service. Appointments will normally be made for the period of one year. Permanent employment opportunities may result in professional status. Annual payment will be \$1,902.00, subject to possible increase by Congressional action. This is a civil service status of SP-4.

Positions are open to three or four qualified graduate students each year who have an abiding interest in the field of mapping. They should have completed an undergraduate major in geography or language and selected one of those fields for a master's thesis or doctor's dissertation.

Students wishing to apply for these positions should send a letter of application with a completed application blank to: E. B. Espenshade, Jr., Chairman, Liaison Committee with Army Map Service, Association of American Geographers, Northwestern University, Evanston, Illinois.

The Research Corporation, a nonprofit organization devoted to advancing research and technology by use of revenues from inventions assigned to it by public-spirited inventors, has announced the initial list of Frederick Gardner Cottrell special grants-in-aid for postwar research, totaling about \$175,000 for the first year of work. The Corporation intends to make grants totaling \$2,500,000 to educational institutions during five years.

Joseph W. Barker, formerly dean of the College of Engineering, Columbia University, and special assistant to the Secretary of the Navy during the war, is president of the Corporation and a member of the Advisory Committee set up to investigate applications for grants. Other members of this committee include W. D. Coolidge, of General Electric Company Laboratories; Thomas N. Chilton, of Du Pont; Lloyd P. Smith, formerly of RCA Laboratories and now of Cornell University; Timothy E. Shea, of Western Electric Company; Col. Staffer Warren, of the Med-

ical School, University of Rochester; and R. R. Williams.

The recipients, projects, and directors, respectively, of the Corporation's initial grants are announced as follows:

Amherst College: design and construction of equipment for measuring faint stars by the photoelectric method, John S. Hall; Boston University: an experimental examination of the properties of the solvated electron by calorimetric, density, and viscosity studies of the alkali-metal, liquid-ammonia systems, Lowell V. Coulter; University of California: the correlation of photochemical processes with molecular absorption spectra, F. E. Blacet; Carnegie Institute of Technology: properties of matter at extremely low temperatures, and superconductivity—especially of thin films, Immanuel Estermann; Catholic University: chemical reactions between gases and solids, Walter John Moore, Jr.; Duke University: microwave absorption spectra of molecules, Walter Gordy; Indiana University: a study of beta disintegration process, L. M. Langer; Kansas State College: study of photoelectric and thermionic properties of spectroscopically and thoroughly outgassed nickel with emphasis on studies at the Curie point, A. B. Cardwell; Michigan State College: ionization produced in gases by electrons of energies less than 2,000 electron volts, Thomas H. Osgood; University of Minnesota: analysis of electron and ion-collision phenomena in gases and vapor, John T. Tate; University of Minnesota: isotopes, A. O. C. Nier; Muhlenberg College: the measurement of the velocities and absorption coefficients of sound waves in gases as a function of temperature and pressure at various supersonic frequencies, I. F. Zartman; University of New Hampshire: a study of inorganic fluorides, H. M. Haendler; North Texas State Teachers College: stereoisomerism of synthetic and natural polyenes, R. B. Escue, Jr.; New York University: investigation of nuclear disintegrations produced by cosmic radiation, Serge A. Korff; University of Notre Dame: (a) extension of experiments on the excitation of nuclei by X-rays and electrons by observation of shorter metastable lifetimes, and (b) extension of experiments on the excitation of nuclei by X-rays and electrons by increasing excitation energy, George B. Collins and Bernard Waldman; Ohio State University: infrared spectra of a large number of polyatomic molecules measured automatically under high dispersion, H. H. Nielsen; Oregon State College: synthesis of amino alcohols derived from pyrimidines and quinazolines, B. E. Christensen; Pennsylvania State College: influence of range of stress on the fatigue strength of metals subjected to axial and biaxial stresses, John A. Sauer; University of Pennsylvania: nuclear physics, G. P. Harnwell; Queens College: isotope research, M. L. Eidinoff; Rutgers University: nuclear and electronic paramagnetism at room and low temperatures, F. G. Dunnington; Stanford University: nuclear induction and its application to polarized neutrons, Felix Bloch; Stevens Institute of Technology: absorption spectra, particularly in the far ultraviolet, of liquids to be

followed later by studies of other properties, etc., E. G. Schneider; Tuskegee Institute: the methods of preparation and the chemical properties of the bis-brominated ethers, C. T. Mason; Southwestern (Memphis): vapor-phase catalytic preparation of high-molecular-weight ketones, J. L. A. Webb; University of Tulsa: distribution of carbon dioxide in systems containing two liquid phases, F. T. Gardner; State College of Washington: (a) enthalpy changes at unit surfaces on wetting of solids by liquids, (b) a study of significance of variations in the values of the densities of fine powders as measured in various liquid media, and (c) development of new techniques for the study of liquid films at solid-liquid interfaces, J. L. Culbertson; and University of Wyoming: determination of neutron resonance energies and half-life periods of induced radioactivities, Emil J. Hellund.

"Research and teaching in our American educational institutions are facing a crisis today," said Dr. Barker. "The general pattern of research in our colleges and universities was seriously disrupted by the war. Talented members of the faculties were drafted into war research or war service. But now the question is whether these men can be brought back to the college laboratories and classrooms where there is a great necessity for them if we are to re-establish adequate research closely associated with inspired teaching."

The Research Corporation was begun in 1912 with the gift, through Frederick Gardner Cottrell, of patent rights on electrical precipitation, used for removing dust, fume, and mists from industrial gases. Over the intervening years the Corporation has served colleges and universities by the administration of patents arising from researches in their laboratories. From the proceeds of these and other patents it has aided in the past some 52 institutions by grants totalling over \$1,250,000. Grants on the normal program are also being continued. In addition, through the Williams-Waterman Fund, grants are being made for research in the combat of dietary diseases.

The Corporation is interested in receiving high-quality applications for grants from engineering and the smaller liberal arts colleges. The address is: 405 Lexington Avenue, New York 17, New York.

Natural Resources of Japan

The Natural Resources Section was established in Tokyo as a special staff section of the General Headquarters of the Supreme Commander for the Allied Powers on 2 October 1945. This section, of which Lt. Col. Hubert G. Schenck (professor of geology at Stanford University, on leave of absence) was appointed chief, was set up to inform and advise the Supreme Commander on all matters pertaining to

agriculture, fisheries, forestry, and mining and geology in Japan and Korea. In order to accomplish its mission, the section is organized into four technical divisions to cover these fields.

Maj. Warren H. Leonard (professor of agronomy, Colorado State College, Ft. Collins, on military leave) was appointed chief of the Agriculture Division and is assisted by Maj. Mark B. Williamson. Lt. Col. Reginald H. Fiedler (chief of the Division of Fisheries Industries, U. S. Fish and Wildlife Service, on military leave) is chief of the Fisheries Division, with Maj. John F. Janssen (marine fisheries biologist, California Division of Fish and Game, on military leave) as assistant. Lt. Col. Arthur R. Spiller (senior forester in charge of State Cooperation, U. S. Forestry Service, on military leave) was named to be in charge of the Forestry Division and, on his return to the United States for discharge from the service, was succeeded by William S. Swingler (assistant regional forester in charge of Division of State and Private Forestry). John J. Collins (formerly assistant regional geologist, Northwest Region, U. S. Geological Survey) was the acting chief in the Mining and Geology Division until the arrival of Thomas A. Hendricks (principal geologist, U. S. Geological Survey) on 6 December 1945 to assume the duties of chief of the Division. Maj. Elmer W. Ellsworth (consulting petroleum geologist) was appointed technical supervisor to coordinate matters involving two or more divisions. Recently Lt. Col. Charles J. Bolner (lawyer, State of New Jersey) joined the staff in order to supervise all activities of the section on Korean affairs.

The major problems with which the section has been concerned up to this time are the provision of commodities essential for sustenance of the Japanese people and for the needs of the occupation forces. Foremost among these problems is the food supply.

The Agriculture Division has been concerned primarily with maximizing the food production in Japan. Activities to implement this objective include an evaluation of the food position; a survey of agricultural research institutions to determine their adequacy for the solution of agricultural problems; a study of the five-year land reclamation program for the addition of 3,828,500 acres to the cultivated area and an analysis of fertilizer requirements to assure agricultural production at a level reached in the 1930's.

Japanese farm lands have been intensively cultivated for centuries, and importation of fertilizer and its raw materials has always been essential. During the war the fertility of the land has diminished greatly. A complete study of sources of raw materials essential for the manufacture of inorganic commer-

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cial fertilizers has been made in cooperation with the Mining and Geology Division. Action is currently under way to provide the raw materials for the production of suitable fertilizers for the use of Japan and other countries in the Orient. These raw materials, together with the large fertilizer-plant capacity of Japan, will do much to reduce the need for the import of large tonnages of food.

Fish constitutes the most important single animal food for the Japanese. Japan has caught more fish than any other nation in the world, but her actions as a fishing nation have been selfish and damaging to international fishing. The Fisheries Division has taken the initiative in helping Japan produce the requirements of a minimum subsistence diet during the immediate postwar emergency. The Division has initiated a program of providing the Japanese fishery fleet limited requirements of fuel oil and fishing equipment. The area opened to Japanese fishing is rigidly prescribed, consisting of waters adjacent to the home islands and a limited area for whaling around the Bonin Islands, although no landings are permitted at that location.

Fuel is next in importance as a commodity necessary to life in Japan. Production of the principal fuel, coal, dwindled almost to the vanishing point with the release of Korean and other slave labor. In addition, mining equipment and mines have deteriorated badly during the war period. The Natural Resources Section initiated the actions that have resulted in raising the monthly coal production to the level of monthly requirements. It is expected that coal production will be further increased.

Charcoal has also long been important in domestic heating in Japan. The Forestry Division has concentrated great efforts on charcoal production and has

done much to meet this demand. This Division is also making a complete study of Japan's forest resources, lumber and allied products, wood pulp, and paper requirements. The forests were severely overcut during the war years, and the lumber and plywood requirements for housing needs of the occupation forces and for reconstruction in Japan are far in excess of the amount of timber that can be supplied domestically without serious damage to the forests and forest lands. All aspects of this problem are being studied in order that it may be resolved as rationally as possible.

In addition to the work on coal and fertilizer the Mining and Geology Division has assembled basic data on all metals, minerals, and petroleum in order to supply information regarding imports essential for the postwar emergency and the export of materials badly needed elsewhere.

The four Divisions have assembled basic data on all natural resources which may be considered as a possible source of reparations. In this connection the Section has received a letter of commendation from the Japanese (Pauley) Reparations Mission for the information, advice, and counsel supplied to that mission.

The Section is also conducting a complete study of Japanese research in the field of natural resources, particularly during the war years. The information obtained in the collection of possible basic data and a study of Japanese research is expected to meet the needs of all current problems. The long-range plan for the work of the Section has been set up with the ultimate goal of obtaining full knowledge of Japanese natural resources for publication as one of more comprehensive reports that should constitute a reference work for many years to come.—*Hubert G. Schenck*, Lt. Col., Chief.

In the Laboratory

Modifications of Specimens in Electron Microscopy

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Ample evidence has been given for the relative stability of many electron-microscope specimens during dehydration and electron bombardment in the instrument. Although a surprisingly large number of specimens maintain their shape, from time to time evidence points to substantial changes of structure or shape

in some objects. The observed stability of specimens under given conditions is by no means proof of similar stability for different specimens or even for the same specimen under changed conditions. Generally speaking, the electron microscopist will have to establish independent proof of the correct reproduction of the structures imaged in the electron microscope before drawing any definite conclusions. Some definite specimen changes are discussed below, and means for their detection and prevention proposed.

For some time during use of the gold shadowing technique of Williams and Wyckoff (3), discrepancies

between the apparent object size and the width of the shadows produced by them were observed. In the case of rather small particles, such discrepancies were not regarded seriously and generally were explained by lack of contrast at the edges of the particles in the unshadowed specimens. Recently, however, such flagrant cases of discrepancy were observed that no such interpretation could be allowed. Fig. 1 reproduces a large particle with a shadow not only considerably wider than the particle but even extending a little



FIG. 1

toward the source in front of the particle. An examination of this and several similar micrographs excluded all interpretation other than a shrinkage of the particle *after* gold shadowing. The specimen in Fig. 1 consists of accidental impurities of unknown origin on a collodion film, which was gold shadowed in an evaporation apparatus and then transferred to the electron microscope. Apparently the particle size changed during observation (focusing) in the electron microscope and shows, therefore, a different appearance from that expected from its shadow. The observed shrinkage in this particular case is as much as 40 per cent. The question whether the particle changed previous to its gold shadowing is left hereby entirely open.

It is proposed, therefore, that in addition to such methods as comparison with other evidence (light microscopy, ultracentrifugation, streaming birefringence, etc.) as much evidence as possible should be gathered in the electron microscope about eventual structural or shape changes during observation. Gold shadowing, if carried out carefully, can be used quite advantageously by comparison of the particle size with the dimensions of its shadow.

Another method of observation is a modification of an earlier procedure outlined by Marton (1), which involved focusing on a dummy specimen and then substituting the true specimen. Von Ardenne (2) applied the same principle by providing a shadowing wedge, protecting part of the specimen during focusing. The modification, as used in this laboratory, consists of carrying out the focusing, which requires a beam of relatively high intensity, on a part of the specimen which can be sacrificed. After the best focus is achieved, the beam intensity is reduced to the minimum required for photographic recording and the stage shifted to a part of the specimen which has not been irradiated previously. A micrograph can then be obtained with a minimum of irradiation and corresponding minimum changes. In a variation of the same procedure the whole surface of the specimen is scanned first at very low intensity and the most interesting part preselected. A shift is then made to a less important area which can be sacrificed. This is brought into focus at high intensity and then, after reduction to low intensity, a shift is made back to the good area. Care should be taken that the two areas are far enough removed so that essentially no high-intensity beam can reach the part which has to be protected. In both cases the filament current can be reduced when the image of the filament is produced in the object plane by means of the condenser lens, and increased again to the optimum value when the condenser setting corresponds to the one required for photographic recording.

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Frozen-dried Preparations for the Electron Microscope

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Most native proteins are strongly hydrated whether they are in molecular suspension or form part of an organized biological tissue. When this water is lost during air-drying for electron microscopy, it is likely that the elementary particles of many proteins will shrink and distort. Dehydration cannot now be avoided, and therefore techniques are needed which will desiccate with a minimum of alteration. Quick-freezing and desiccation from the frozen state does this.

Good frozen-dried preparations for electron microscopy can be made easily in the following way: Aluminum strips are substituted for glass microscope slides in mounting the usual collodion- or formvar-covered screens. When ready for use, these metal slides and the screens they carry are placed on a block of metal (approximately $8 \times 3 \times 2$ cm.), precooled with dry ice or liquid air. When a microdrop of solution is applied to, and immediately withdrawn from, screens cooled in this fashion, some will instantly freeze; a tissue such as tendon, touched momentarily to a cold screen, will leave frozen shreds behind. The collodion or formvar substrate can often be omitted when dealing with tissues and with suspensions of elongated particles. The block has sufficient thermal capacity to hold the preparations frozen while they are being made, while both block and preparations are being transferred to a vacuum chamber, and until a high vacuum has been drawn. If the preparations are to be shadow-cast before microscopic examination, vacuum desiccation and shadowing can conveniently take place in the same apparatus without breaking vacuum. The screens on their metal block must remain well below freezing until desiccation has been completed, but by the end of the run they should be warm enough so that moisture from readmitted air does not condense on them. The slow thermal leakage

this demands is provided by putting the block on one or more pieces of lightly metal-coated glass.

Electron micrographs have thus far been made of frozen-dried bacteria, of several plant and animal viruses, and of certain tissues. With dilute solutions of the tobacco mosaic and bean mosaic viruses, for example, these pictures are indistinguishable from those of ordinary air-dried preparations; in such instances it would appear that air-drying does not appreciably distort the virus particles. Influenza virus particles have been strikingly full and turgid after freeze-drying. Unusually interesting pictures have been obtained from frozen-dried concentrated solutions of tobacco mosaic protein. In them many rods are associated together in two dimensions to yield areas which look surprisingly like sheets of connective tissue and break up in the same way into fibrous bundles—for example, under impact of the electron beam. Frozen-dried preparations containing appreciable quantities of salt have not yet given useful micrographs, because in its extreme dispersion this salt tends to smear over the fine details that are present.

In this laboratory frozen-dried as well as air-dried preparations are now a routine. Electron micrographs of some of them will shortly be published elsewhere.

Letters to the Editor

On the Mechanism of Action of Folic Acid and Liver Extract in the Treatment of Anemia

In view of the discovery by Spies and co-workers (*S. med. J.*, 1945, 38, 707) that synthetic folic acid has antianemic action in the treatment of human macrocytic anemias, the question of its mechanism of action has become a matter of some interest. Experiments performed in our laboratory (details to be published) may throw considerable light on this problem.

We have produced significant hyperchromic anemias in five normal dogs by the subcutaneous injection of 3 mg. of acetylcholine bromide twice daily for 47 days. Two of these dogs were then treated by the daily injection of liver extract, in addition to acetylcholine. They responded with an increase of reticulocyte percentage and a gradual regeneration of red blood cells to their normal number. Another dog of this series received daily folic acid injections (2 mg.) and responded in a similar manner.

Three dogs were made anemic by the feeding of choline chloride according to the general method reported previously by the author (*Amer. J. Physiol.*, 1944, 142, 402).

Two of these dogs were treated with daily injections of folic acid and the third, after serving as an anemic control animal, was treated daily with liver extract. These animals all responded by showing a rise of reticulocytes (to peaks of 3.4–4.2 per cent, from 6 to 9 days after onset of treatment) and a return to normal of their erythrocyte numbers within 20 days, in spite of continued choline feeding.

During anemia, acetylcholine-like activity was detected in extracts of serum of blood drawn from the "choline anemia" dogs at one and one-half hours after the administration of 200 mg. of choline chloride. This activity was markedly diminished after antianemic treatment had been instituted. Cholinesterase activity (determined by an electrometric titration method) of the serum of one dog tested was low during anemia and was increased 12-fold during treatment with liver extract.

Incubation of various dog blood sera with folic acid or liver extract at 37°C . increased their cholinesterase activities by from 0 up to 93 per cent. Similar incubation of one normal human serum with liver or folic acid increased its activity by 15 per cent.

Oral administration of 5-7.5 mg. of folic acid to two normal human subjects increased their serum cholinesterase activities by 33 and 16 per cent within five hours.

It is concluded from these experiments that liver extract and folic acid act by increasing, in some manner, the formation of cholinesterase in the body.

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Some Effects of Electronic Transitions Upon Precision Thermometry

Recent measurements of the electrical resistance of various materials at elevated temperatures have disclosed the following information which may be of value in precision thermometry:

(1) The electrical resistance of a pure conductor is a straight-line function of temperature, but the slope changes appreciably between certain specific temperatures.

(2) Since the temperatures at which these discontinuities have been found are independent of purity, concentration, or heat treatment, the resistance-temperature curve for an alloy will be affected to some extent at each of the temperatures which are characteristic of each of its components. These temperatures may be used as an accurate method of calibration in the proper temperature range. The discontinuities in the curve for carbon, for example, are particularly satisfactory for calibration in the range above the melting point of gold.

(3) Errors may be introduced in certain ranges of temperature by the common practice of drawing calibration curves smooth instead of as straight lines changing in slope at these specific temperatures. These errors may be as large as 6° C. in a chromel-alumel thermocouple or as large as 2° C. in a platinum resistance thermometer. The chromel-alumel thermocouple is free from these errors below about 160° C., and the platinum thermometer is not affected markedly except in the range 160°-932° C., most of the trouble being between 160° and 800° C.

(4) Heat treatment is equally as important as purity in affecting the temperature coefficient of resistance for platinum. Depending upon the heat treatment, values of the coefficient as high as 1.400 or as low as 1.366 have been secured, using the same specially prepared high-purity wire.

Further information on these points will be published in the near future.

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Some Thoughts on "Gene Action"

Dr. Deakin's recent letter (*Science*, 1946, 103, 570-572) prompts me to add some thoughts which I noted down some time ago on the same subject. I am presenting them merely in the hope that they may invite extended discussion of this problem.

It has been generally accepted that the control of hereditary factors is closely associated with the desoxyribonucleic acid (DRA)—protein components of the chromosomes. A widely held concept has attributed to these

components the ability of catalyzing enzyme processes and has assigned to them a principal role in the production of enzymes. In view of some recent work, however, the question may be raised whether all action (or some action) of DRA may not be in the nature of *inhibiting* enzyme processes, either qualitatively or quantitatively. Among the recent work to which this might be applied are the data by Avery and co-workers on the specific transformation of bacterial types of DRA, the data by Dickinson on the suppression of bacterial mutation through enzyme inhibitors, as well as the results of Lindgren on yeast and Sonneborn on *paramecium*. Furthermore, *in vitro* tests by Greenstein have actually demonstrated the ability of DRA to inhibit enzyme reactions. If the action of DRA in the chromosomes is totally or partly of an inhibiting nature, it implies that the extra-chromosomal material contains many more ultimately possible enzyme reactions than those actually realized during the development and life of an organism, since many of them would be blocked by the chromosomal constituents. This block may or may not be a total one. The inhibition may be in some cases a quantitative one, delaying time and amount of action of a particular enzyme. A loss in DRA would result in the release of one or more additional enzymatic processes. This possibility may be realized in the well-known mutation due to chromosomal deficiencies. This concept would assign a much greater importance to extrachromosomal constituents than has hitherto been customary. Extrachromosomal constituents as long as they remain stable, would limit the extent of variation possible through changes in the chromosomes since the ultimately possible enzyme reactions would then be exhausted if none of them is blocked by chromosomal constituents (microevolution). However, changes or extrachromosomal constituents may occur, but far less frequently than changes of chromosomal constituents. Such changes would then permit the realization of completely different enzyme processes, dependent on the extent of their quantitative or qualitative inhibition by chromosomal constituents (macroevolution).

Incidentally, the inhibiting-factor hypothesis is not altogether a new concept. Bateson, for example, speculated along these lines as early as 1913 (*Problems of genetics*. Yale Univ. Press, esp. pp. 94-96).

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Meteor Crater, Arizona

In December 1945 Nelson H. Darton restated to the Geological Society of America, and also to the Association of American Geographers, his belief that Meteor Crater east of Flagstaff on the Arizona Plateau, is of volcanic rather than meteoritic origin. He cited a decision of the U. S. Board on Geographic Names in which the name *Crater Mound* was officially adopted, and he urged that the use of the term *Meteor Crater* be discontinued. Since notices of Mr. Darton's views have been published in *Science News Letter* and other nontechnical media, seems timely to indicate that the majority of geologists

astronomers, engineers, and others, who have studied the crater disagree with Mr. Darton and are convinced that the crater was made by the impact of a large meteorite.

Mr. Darton appears to stake his opinion largely on the failure of exploration (by drilling and geophysical methods) to reveal the presence of a buried meteorite beneath the crater. He is perhaps not aware that in 1930 F. R. Moulton showed that if a large meteorite did strike the plateau, it must have developed at the point of impact such a high temperature as to result not only in a tremendous explosion but also in vaporization of the meteorite itself, along with part of the surrounding rock strata. Under those circumstances one could not expect now to find more than incidental fragments of the meteorite.

Mr. Darton also seems to ignore the significance of the

unique composition of the parapet which surrounds the crater and the material which partially fills the cavity itself. He is perhaps unaware that underneath the surface rubble these consist largely of quartz powder and silica glass, derived from the underlying sandstone by pulverizing and melting. No materials of this kind have ever been found in association with volcanoes, and temperatures high enough to produce silica glass are probably rarely, if ever, attained in volcanic eruptions.

It seems, therefore, that the current use of the name Meteor Crater is well justified, and the field evidence is heavily against the hypothesis of volcanic origin.

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Book Reviews

Encyclopédie entomologique. XXII: Les Coléoptères des denrées alimentaires et des produits industriels entreposés. P. Lepesme. Paris: Paul Lechevalier, 1944. Pp. 335. (Illustrated.) 350 fr.

This is a compilation of information useful and valuable to the general worker and to those interested in coleopterous insects injurious to stored products. It should also prove of interest to those in other fields of work. However, a specialist may find his particular field inadequately treated and will probably disagree with the author on certain points.

The work is divided into two parts. Part I deals primarily with the taxonomy and descriptions of certain species of 24 families, with keys to the families and to the species discussed. Common names in one or more languages are given in addition to the scientific name. The specific descriptions are too brief for general taxonomic purposes, but they are given in sufficient detail for the purpose of this paper. In the case of most species the author has included data on geographic distribution, biology and damage, life cycles, and natural enemies. A total of 214 figures illustrate Part I.

Part II deals with theory and general information concerning the beetle population of stored products. In it are considered and discussed the environment of the food products, including constant and variable factors; the relationship or bond between the insects and the products; diet and the climatic factor; geographical distribution, broadly but briefly treated; life cycles and factors influencing them; the relationship of insects with other organisms; the theory of "vacant space" and the coleopterous population of the food products; tropisms; hybridization and variation; biological equilibrium; damage; and means of control. Because of the scope of each subject covered, it is obvious that only the essentials

could be mentioned. It appears, therefore, that it was the desire of the author to expose the reader, however briefly, to some of the factors influencing insects and to some facts and information that should be known by the general worker for a clearer understanding of, and a better approach to, the control of insects infesting stored products. The section is illustrated with 19 figures, one of which is a diagram for a fumigator in which methyl bromide is employed.

The paper is terminated with an extensive bibliography, an index to genera and species, an index of common names, and 10 plates of commendable photographs demonstrating the damage caused by various insects. Plates 11 and 12 contain reproductions of photographs of installations for the fumigation of the various products.

If the purpose of the paper is correctly interpreted, it would have been greatly improved and made more practical if, in some instances, more detailed information had been given for the individual species and not reserved for a general discussion of a closely related group of species. This paper and the one by Hinton (1945) supplement each other.

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A primer of electrocardiography. George Burch and Travis Winsor. Philadelphia: Lea and Febiger, 1945. Pp. 215. (Illustrated.) \$3.50.

The reviewer was disappointed in his hope that this book by Burch and Winsor might be the long-awaited book for medical students and those beginning the study of electrocardiography. It is timely and has much to recommend it, but it has many faults.

One thinks of a primer as a book with which to begin and as embodying the simplest ideas, with the notion of a gradual progression by means of other books into the more difficult phases of a subject. This is not the case with this book. It deals with the most complex and theoretical sides of electrocardiography. Neither the medical student, who has not the time for this kind of background in electrocardiography—a rubric which must take up a small part of the total time spent in medicine in relation to the other branches of his courses—nor the physician, having only a clinical interest in electrocardiograms, would be anything but hopelessly lost and discouraged by this introduction to electrocardiography. A title which would foretell its real contents would be more satisfactory and not misleading.

There seems to be no occasion for, or purpose in, drawing out so laboriously diagrams of the time lines and lines for amplitude and diagrams of the galvanometer string movements, when actual electrocardiograms with the actual photographed lines could have been used and enlarged if it was thought contributory to the clearness of demonstrating time intervals, elevations of segments, etc. Certainly a reality would have been achieved which the diagrammatic fashion misses. Moreover, when the lines are not accurately drawn equidistant or parallel, one is very conscious of these defects. Moreover, from the point of view of reading and study, the drawing of time lines and voltage lines only immediately surrounding the movement of the galvanometer string is annoying, distracting, and highly artificial.

The reviewer could not find anywhere in the book a reproduction showing what an electrocardiogram actually looks like when taken for clinical purposes. Anyone reading this primer as an introduction would come away with the idea of the cross lines being written only in the field surrounding the electrocardiogram string movement.

Some of the diagrams indicating starlike explosions in the heart detract from the seriousness and depth of the rest of the book.

The assets of the book must be considered in the light of the defects listed above.

The introductory chapter on the theory of the electrocardiogram is good, as are the text relating to the analysis of the waves of the electrocardiogram and over-all analysis of the electrocardiogram, and the chapter on precordial chest leads.

The data relating to myocardial infarction should be brought together in one section instead of being spread out in several parts; for instance, there are descriptions on page 94 and again on page 170. It does not seem wise however, to teach the idea of acute, subacute, and chronic myocardial infarction with the connotation of these words in medicine.

The chapter on "Disorders of the Heart Beat" is not very effective and leaves much to be desired for recommendation to medical students and physicians.

The most useful function this book serves is to bring together in a compact way data relating to the mono-

cardiogram, vector analysis, and ventricular gradient pointing up the work of Mann, Wilson, Ashman, and Bayley, and the earlier German work. The authors have made a great effort to stress these investigations. This section is well done; at the moment, however, for clinical electrocardiography these analyses have not a great deal of usefulness, and their eventual clinical value is not yet clear. Accordingly, in Chapter V, "The Clinical Applications of the Electrocardiogram," this phase is overemphasized for the state of its importance at the present time.

On page 189, relating to the "Diagnostic Value of the Electrocardiogram," the authors speak of electrocardiographers and clinicians. The reviewer hopes that in medicine there will never be anyone who is called an "electrocardiographer." No one should read or interpret electrocardiograms who is not interested actively in clinical medicine and who is not a clinician.

Much of the data in the Appendix has usefulness. Carter's chart for measuring the angle of the electrical axis might have been included. A list of references also would have been a valuable addition to the book.

This book is not for the beginner or for the medical student, but only for those who, having had an introduction to electrocardiography by other means, wish to go further into the subject.

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The mosquitoes of New Jersey and their control. Thomas J. Headlee. New Brunswick, N. J.: Rutgers University Press, 1945. Pp. x + 326. (Illustrated.) \$4.00.

This is an enlarged and revised edition of Bulletin 348 of the New Jersey Agricultural Experiment Station, published in 1921 under the same title and long a standard reference for mosquito workers of the United States.

The present volume opens with three short chapters on the "Value of Mosquito Control," "Structure, Classification, and Keys," and "The New Jersey Mosquito Fauna." By far the largest section (200 pp.) is entitled "Mosquito Biology" and includes technical descriptions and illustrations of the adults and larvae of the species found in New Jersey. As stated by the author, the bulk of the material in this chapter has been taken directly from the 1904 report of John B. Smith, a notable pioneer in this field. His report has been out of print for many years, and its reproduction in this manner is of undoubted historical value. It would seem, however, that briefer summaries of the pertinent facts, now well established, would fill present-day needs better than the details of the original observations and experiments. Very little new information has been added, except for records of light-trap collections from 1932 to 1941, and one must look in vain in most cases for an account of the present status of the different species or information that has been accumulated during the intervening 40 years. Bulletin 348 and other publications must be referred to for information of this nature. There is evidence that this chapter has not been read critically with respect either

to certain details of bionomics and morphology or to bringing the information up to date. For example, after giving Dr. Smith's observations on the hatching of salt-marsh mosquito eggs, no reference is made to more recent discoveries of the factors that influence hatching. Taxonomic characters of more value than those provided are frequently omitted, and the subject matter on the different species is somewhat unevenly presented.

In a short chapter (18 pp.) on "The Principles and Detailed Procedure of Mosquito Control," the author summarizes, all too briefly, the subject on which he can speak with the greatest authority, having led the work in New Jersey for 30 years or more. His nine basic principles and his opinions on salt-marsh ditching are of great value. A short chapter on "Larvicides" is devoted chiefly to pyrethrum extract emulsion, as developed in New Jersey, and its use in the protection of outdoor gatherings against adult mosquitoes. Petroleum oils and iron or copper sulphate are the only other materials mentioned. No consideration is given to recent developments in the use of DDT.

The remaining chapters cover the subjects of environment, history of mosquito control in New Jersey, mosquito repellents, laws relating to mosquito control, and the economic effect of mosquito reduction. There is a short bibliography of 30 titles, most of which are of early work.

While the volume is local in content and little attention is given to methods employed or work done elsewhere, there is considerable material of general interest.

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Table of arc sin x and Tables of associated Legendre functions. Lyman J. Briggs, Arnold N. Lowan, et al. (Prepared by the Mathematical Tables Project, conducted under the sponsorship of the National Bureau of Standards.) New York: Columbia Univ. Press, 1945. Pp. xix + 121; xlv + 306. \$3.50; \$5.00.

These are two further welcome volumes of the series prepared under the auspices of the WPA of New York City and the OSRD.

The first of these volumes tabulates arc sin x to 12 decimal places, for values of the argument differing by 0.0001, and to permit interpolation, tabulates also second differences; for x near unity this plan is somewhat modified to insure more accurate interpolation. Auxiliary tables are provided for convenience in the use of the volume with interpolation.

The second of these volumes tabulates the functions $P_n^m(x)$, $Q_n^m(x)$, and their first derivatives, for integral and half-integral values of n, integral values of m, and real and pure imaginary values of x, and also tabulates the functions $P_n^m(\cos \theta)$ and their first derivatives for integral values of n and m. In the main, tabulations are to six significant figures, for $1 \leq n \leq 10$, $0 \leq m \leq 4$, and $1 \leq x \leq 10$. Tabulation of the derivatives mentioned is especially noteworthy, having been largely neglected in previous tables.

The present volumes are highly useful tools in the current development of mathematics in the direction of nu-

merical computation and physical application. The sponsor, Dr. Briggs, and the director, Dr. Lowan, are again to be congratulated on their planning, preparation, and publication.

J. L. WALSH

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The university at the crossroads. Henry E. Sigerist. New York: Henry Schuman, 1946. Pp. viii + 162. \$2.75.

It is much the fashion these days to write about university education. Many more people are interested, since many more people are concerned than was the case even a few years ago. In addition, the furor over the peculiar Thomistic revival at Chicago has excited much argument over the purposes and methods of university education.

Dr. Sigerist has been disturbed by the impact of the current cultural revolution, as exemplified by the war, on university, and particularly on medical, education. The title essay of the 12 included in this small volume was written in 1944 for the *Bulletin of the History of Medicine*, in which eight others of the essays were first printed. Two, "Failure of a Generation" and "The Social Sciences in the Medical School," are published for the first time in this volume.

A collection of essays of this sort is bound to be uneven, but all illustrate well Dr. Sigerist's crusading spirit. While there is a strong pessimistic tone to most of the essays, it is applied chiefly against the quite well-known defects of current university education in the United States. Dr. Sigerist seems to think that all will be well if university professors and university-trained people will take a more active part in governmental and social affairs.

Much interesting autobiographical material is offered in the essay entitled "University Education," delivered in 1939 at Johannesburg. In this address, Dr. Sigerist reveals his interest and his prejudices. He also indicates the various men and circumstances which have so profoundly influenced him.

Five of the 12 essays deal with medical educational problems, but in a manner illustrating the way by which medical education may be correlated with a general cultural training. Dr. Sigerist pleads vigorously for an appreciation of the classic contributions on which our culture rests and urges an extension of research into their social and cultural applications.

As director of the Institute for the History of Medicine at The Johns Hopkins University, Dr. Sigerist is in a strategic position to influence cultural trends, particularly with reference to one of the great professions. He need not be disappointed at the relatively slow acceptance of his ideas and proposals. On the contrary, he may take much satisfaction in his own stimulating contributions and in the solid achievements of his many pupils. He is himself exemplifying the reasonable sort of a path which the university may profitably take when it finds itself at a crossroads.

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Scientific Book Register

- BELL, CLIFFORD, and THOMAS, TRACY Y. *Essentials of plane and spherical trigonometry*. (Rev. ed.) New York: Henry Holt, 1946. Pp. ix+163. \$2.30 with tables; \$2.00 without tables.
- BROOM, ROBERT, and SCHEPERS, G. W. H. *The South African fossil ape man: the Australopithecinae*. (Transvaal Museum Mem. No. 2.) Pretoria, Union of South Africa: Transvaal Museum, 1946.
- BURKET, LESTER W. *Oral medicine*. Philadelphia: Lip-pincott, 1946. Pp. 674. (Illustrated.) \$12.00.
- CHRISTENSEN, CLYDE M. *Keys to the common fleshy fungi*. Minneapolis: Burgess Publishing Co., 1946. Pp. 45. \$1.50.
- COLBERT, EDWIN HARRIS. *The dinosaur book; the ruling reptiles and their relatives*. (Man and Nature Publ., Handb. No. 14.) New York: American Museum of Natural History, 1945. Pp. 156. (Illustrated.) \$2.50.
- COMMITTEE FOR THE CALCULATION OF MATHEMATICAL TABLES. *Mathematical tables*. Vol. I: *Circular & hyperbolic functions, exponential & sine & cosine integrals, factorial function & allied functions, Her-mitian probability functions*. (2nd ed.) Cambridge, Engl.: At the Univ. Press, 1946. Pp. xi+72. \$2.50.
- DICKEY, GEORGE D., and BRYDEN, CHARLES L. *Theory and practice of infiltration*. New York: Reinhold, 1946. Pp. v+346. (Illustrated.) \$6.00.
- DUNCAN, JOHN CHARLES. *Astronomy: a textbook*. (4th ed.) New York-London: Harper, 1946. Pp. vii+500. (Illustrated.) \$4.50.
- EDELSTEIN, EMMA J. and LUDWIG. *Asclepius: a collec-tion and interpretation of the testimonies*. Baltimore: Johns Hopkins Press, 1945. Vol. I: Pp. xvii+470; Vol. II: Pp. x+277. \$7.50.
- EGLOFF, GUSTAV. *Physical constants of hydrocarbons*. New York: Reinhold, 1946. Pp. xiii+661. (Illus-trated.) \$15.00.
- FARADAY, JOSEPH E. (Comp.) *Encyclopedia of hydro-carbon compounds*. Brooklyn, N. Y.: Chemical Pub-lishing Co., 1946. \$15.00.
- FISHBEIN, MORRIS. *The popular medical encyclopedia*. Garden City, N. Y.: Doubleday. Pp. 540. (Illus-trated.) \$4.95.
- GREENHILL, J. P. *1945 year book of obstetrics and gynecology*. (Practical Med. Ser.) Chicago: Year Book Publishers, 1946. Pp. 576. (Illustrated.) \$3.00.
- HAMBLY, WILFRID D. *Craniometry of Ambrym Island*. (Fieldiana: Anthropology, Vol. 37, No. 1.) Chicago: Chicago Natural History Museum, 1946. Pp. viii+150. (Illustrated.) \$2.75.
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- HELDMAN, JULIUS D. *Techniques of glass manipulation in scientific research*. New York: Prentice-Hall, 1946. Pp. xii+132. (Illustrated.) \$3.60.
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- PIWOWARSKY, EUGEN. *Hochwertiges Gusseisen, seine Eigenschaften und die physikalische Metallurgie seiner Herstellung*. Ann Arbor, Mich.: J. W. Edwards, 1946. Pp. 1014. (Illustrated.) \$28.50.
- PRESCOTT, SAMUEL CATE; WINSLOW, CHARLES-EDWARD A.; and MCCRADY, MAC HARVY. *Water bacteriology with special reference to sanitary water analysis*. New York: Wiley; London: Chapman and Hall. Pp. 366. \$4.50.
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- VON BRAND, THEODOR. *Anaerobiosis in invertebrates*. Normandy, Mo.: Biodynamica, 1946. Pp. 328. \$4.80.
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